MONOSODIUM GLUTAMATE VERSUS DIET INDUCED OBESITY IN PREGNANT RATS AND THEIR OFFSPRING

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ABSTRACT

Background and aim: We aim to determine the role of MSG compared with high caloric chow (HCC) in development of obesity in pregnant rats and their offspring.

Materials and Methods: Ninety pregnant rats were divided into 3 groups, control, MSG and HCC fed. We determined body weight (BW), body fat, fat to body weight ratio, serum glucose, insulin, leptin, lipid profile, ob and leptin receptor-b gene expressions in pregnant rats and ob and leptin receptor-b gene expressions, serum insulin, glucose, leptin, triacylglycerides, total lipids and body weight in offspring.

Results: Daily food intake and BW decreased while body fat and fat body weight ratio increased in MSG relative to HCC fed rats. MSG or HCC increased Ob gene expression, leptin, insulin, LDL, VLDL, cholesterol, total lipids (TL), glucose and decreased leptin receptor-b gene expression. In offspring of MSG treated rats, BW, serum glucose, insulin, leptin, triacylglycerides, TL and Ob gene expression increased and leptin receptor-b gene expression decreased whereas in offspring of HCC fed rats, serum insulin, leptin, Ob and leptin receptor-b gene expression increased but no significant increase in serum glucose, triacylglycerides, TL or BW.

Conclusion : We conclude that in pregnant rats, MSG, in spite of mild

hypophagia, caused sever increase in fat body weight ratio, via leptin resistance, whereas, HCC increased BW and fat body weight ratio, due to hyperphagia with consequence leptin resistance. Moreover, maternal obesity in pregnancy, caused by MSG, has greater impact on offspring metabolism and BW than that induced by HCC.

Key words : obesity, pregnant rats, monosodium glutamate, high caloric chow.

INTRODUCTION

The global epidemic of obesity and associated diseases has a major impact on human morbidity, mortality and quality of life, and is a major drain on healthcare resources [1, 2]. Obesity in pregnancy represents a special problem not only because of adverse effects on maternal health and pregnancy outcome [3] but also because of growing evidence for persistent and deleterious effects on the developing child [4, 5]. With all the attendant complications of obese pregnancy (hypertensive disorders, gestational diabetes, thromboembolic events and Caesarean section), the increased cost of prenatal care alone in obese women can be as much as 16-fold compared with normal weight subjects [6], and the added cost of neonatal intensive care for infants compromised by these disorders also needs to be considered. However, it is the potential social and economic cost in terms of the health of future generations that may present the biggest burden.

The etiology of obesity is multifactorial and is becoming a problem of public health, due to its increased prevalence and the consequent repercussion of its co morbidities on the health of the population. Obesity can be induced in animals by neuroendocrinal, dietary or genetic changes. The most widely used models to induce obesity in rats are by inducing a lesion of ventromedial hypothalamic nucleus by the administration of monosodium glutamate (MSG) or a direct electrical lesion, ovariectomy, feeding on hypercaloric diets and genetic manipulation [7].

Postnatal subcutaneous MSG administration permanently damages the hypothalamic arcuate nucleus

producing obesity [8] and endocrinological dysfunctions as decreased pituitary growth hormone, due to impairment of production of growth hormone releasing hormone, and luteinizing hormone which results in shorter body length, atrophied pituitaries, gonads, and optical nerves [9]. On the other hand, diet induced obesity has a late onset and was developed after feeding mice with high energy diet. It is similar to common human obesity resulting from overconsumption of food and sedentary style of life. It is characterized by leptin and insulin resistance [8].

This work was designed to study the effect of oral administration of prolonged large doses of MSG and high caloric chow on ob and leptin receptor-b gene expression, lipid profile and its controlling hormones and their relationship to obesity in pregnant rats whilst studying the health status of their offspring.

MATERIALS AND METHODS

Animals

Ninety female albino rats, weighting 150±20g each, were purchased from lab animal farm (Helwan, Cairo, Egypt) and housed (Animal House, Medical Physiology department, Faculty of Medicine, Mansoura University, Egypt) in standard cages in groups of four to six animals per cage under controlled conditions (temperature 25±0.5°C, a 12:12 light/dark cycle), with free access to food and water. Food intake over 24 hours was estimated by measuring the amounts of each food consumed. Body weights were recorded regularly throughout the experiments. Body fat and fat to body weight ratio were also detected. All experimental procedures of the present study were approved by the Medical Research Ethics Committee of Mansoura University, Egypt.

Rats were randomly divided into three groups :

Group (I) : Control (C) group (n = 30) fed Purina rat chow (no.5001) ad libitum, which contains 3.3 kcal/g with 23.4% protein, 4.5% fat, and 72.1% carbohydrate which is primarily in the form of complex polysaccharide [10].

Group (II) : Monosodium glutamate (MSG) Group (n = 30): fed

Purina rat chow(no.5001) ad libitum, supplemented with 100 mg MSG (Sigma, St. Louis, MO) per Kg of rat body weight according to Diniz et al [11].

Group (III) : High caloric chow Group (n = 30): fed high caloric chow (HCC) (no. C11024F, Research Diets, New Brunswick, NJ), which contains 4.47 kcal/g with 21% of the metabolizable energy content as protein, 31% as fat, and 48% as carbohydrate,50% of which is sucrose [10].

The three groups were kept in the same conditions for three months. Sub-sections of the three groups were allowed to mate whilst continuing on the same dietary regimens. Then, each of the three pregnant groups was further classified into three subgroups.

Group (I) : Control (C) pregnant group (n = 30): was further classified into three subgroups:

Subgroup Ia: control pregnant rats (n = 10) sacrificed by decapitation at 10th day of pregnancy (C_{10}).

Subgroup Ib: control pregnant rats (n = 10) sacrificed by decapitation at 20th day of pregnancy (C_{20}).

Subgroup Ic: control pregnant rats (n = 10) allowed to go into labour then their male offspring were sacrificed by decapitation at 10th post natal day of age (PN_{10}-C).

Group (II) : Monosodium glutamate (MSG) pregnant Group (n = 30): was further classified into three subgroups:

Subgroup IIa: MSG treated pregnant rats (n = 10) sacrificed by decapitation at 10th day of pregnancy (MSG_{10}).

Subgroup IIb: MSG treated pregnant rats (n = 10) sacrificed by decapitation at 20th day of pregnancy (MSG_{20}).

Subgroup IIc: MSG treated pregnant rats (n = 10) allowed to go into labour then their male offspring were sacrificed by decapitation at 10th post natal day of age (PN_{10}-MSG).

Group (III) : High caloric chow preg-

nant (HCC) Group (n = 30): was further classified into three subgroups:

Subgroup IIIa: high caloric chow fed pregnant rats (n=10) sacrificed by decapitation at 10th day of pregnancy (HCC_{10}).

Subgroup IIIb: high caloric chow fed pregnant rats (n=10) sacrificed by decapitation at 20th day of pregnancy (HCC_{20}).

Subgroup IIIc: high caloric chow fed pregnant rats (n = 10) allowed to go into labour then their male offspring were sacrificed by decapitation at 10th post natal day of age (PN_{10}-HCC).

Sampling protocol

Blood samples At the end of experimental period, rats from all groups were sacrificed by decapitation and blood samples were collected without anti coagulant to obtain serum for determination of serum glucose, insulin, leptin, lipoprotein lipase activity(LPL), apo A1, apo B, triacylglycerides, cholesterol, high density lipoproteins (HDL), low density lipoproteins (LDL), very low density lipoproteins (VLDL), total lipids (TL).

Tissue samples : Abdominal fat pads and hypothalamus were dissected rapidly, post-mortem and kept in liquid nitrogen until used for determination of ob and leptin receptor-b gene expression respectively.

Methods

Determination of ob and leptin receptor-b gene expression using a semi-quantitative RT-PCR according to Meadus [12].

Total RNA preparation.

Hypothalamus and abdominal fat bad samples were pulverized in liquid nitrogen and total RNA was prepared from the frozen powder using the E.Z.N.A ^{TM}.spin column RNA extraction kit (Omega Bio-Tech, Cat NO R6834-01, Canada) following the manufacturer instructions.

The amount of extracted RNA was quantified by measuring the absorbance at 260 nm. The purity of RNA was checked by the ratio between the absorbance values at 260 and 280

nm and ranged between 1.8 and 2.1, demonstrating the high quality of the RNA. This was confirmed in 52 (16%) randomly selected samples by electrophoresis on 1.5% agarose gel containing ethidium bromide.

RT-PCR. One-half microgram of total RNA was denatured for 10 min at 72 °C and was reversed transcribed to cDNA by incubating with 10 µl RT reaction mixture containing: 50 mM Tris-HCl, 75 mM KCl, 3 mM $MgCl_2$, 10 mM dithiothreitol, 0.5 mM of each dNTP, 62.5 mU RNA guard, 50 ng random hexamers and 100 U Superscript II reverse transcriptase. Incubation was performed at 42 °C for 60 min, heated to 95 °C for 5 min, and then quickly chilled on ice.

The PCR reaction mixture contained 2 µl cDNA, 0.2 mM of each dNTP, and the Taq polymerase buffer, which contained 10 mM Tris-HCl pH 8.3, 50 mM KCl, 1.5 mM MgCl2, 7.5 pM of each primer and 1.5 U of Taq polymerase.

PCR conditions were a denaturation step at 95 °C for 2 min followed by 28 cycles of 95 °C, 1 min; 55 °C, 1 min; 72 °C, 1 min. PCR were performed with a 2720 thermocycler (Applied Biosystems, USA). PCR products were analyzed on a 2% agarose gel in 90 mM Tris-borate, 2 mM EDTA buffer (TBE), pH 8, and visualized by staining with ethidium bromide and UV transillumination, band intensity was measured with a Storm 840 Phosphor Imager and quantified with Image Quant software (Molecular Dynamics).

Controls without reverse transcriptase were systematically performed to detect any cDNA contamination.

Primers. Primer design was optimized for multiplex polymerase chain reaction (PCR) with EugeneTM version 2.2 (Daniben Systems, Cincinnati, OH) using the following restrictions: oligonucleotide length of 18-25 bases, GC content of 30-70%, melting temperature close to 60 °C, product length within 150-650 bases, and maximum 6-8 primer dimer formation allowed within multiplex PCR.

In this semiquantitative RT-PCR method, three different primers sets

were used [13] one primer set used for the rat ob gene had the following sequences: sense 5'-GAC ACC AAA ACC CTC ATC AAG-3' and antisense 5'-ATG TCC TGC AGA GAG CCC TG-3'. With this primer set, PCR generated a 383-bp fragment (GenBank accession no, NM 013076).

The second primer set was specific for rat Ob-Rb forward (5'-TGA CCA CTC CAG ATT CCA CA-3') and reverse (5'-AAG CTC ATC CAA CCC TGA GA-3'), yielding a product size of 350-bp (GenBank accession no, D85558).

The third primer set was specific for rat TATA-box binding protein (TBP) which is an ubiquitously housekeeping gene and can thus be used as internal standard. Rat TBP gene specific primers had the following sequences: sense 5'-ACC CTT CAC CAA TGA CTC CTA TG-3' and antisense 5'-ATG ATG ACT GCA GCA AAT CGC-3'. With this primer set, PCR generated a 190-bp fragment of the TBP gene.

Biochemical investigations :

Serum total lipids were performed by colorimetric method using sulfo-phospho-vanilic miture (kit obtained from SPINREACT,S.A.U Ctra.Santa Coloma, Spain, Ref: 1001270). Plasma TG concentrations were measured by the peroxidase method using a commercial kit (Ref 41030, SPINREACT, Spain). Serum total cholesterol was performed using the cholesterol oxidase method where the pink color of quioneimine was measured at 500 nm The used kit was supplied by SPINREACT, Spain (Ref: 41020). Plasma HDL cholesterol was determined enzymatically after precipitation of apoB-containing lipoproteins by the phosphotungstic acid-Mg method with the kit supplied by SPINREACT,Spain (Ref: 1001095). Plasma LDL cholesterol was estimated, after measuring of TAG and HDL, according to Friedewald Formula (14): LDL cholesterol = Total cholesterol - (Triglycerides/5 + HDL cholesterol). VLDL was calculated from TAG according to Friedwald Formula (14), VLDL = TAG / 5. Plasma glucose was quantitated by glucose oxidase-

peroxidase method using the kit supplied by SPINREACT, Spain (Ref:1001190). Serum insulin and leptin were determined using Ultra Sensitive Rat Insulin ELISA Kit (Cat.No. 90060, Crystal Chem INC, Spain) and Rat Leptin ELISA Kit (Cat.No. 90040, Crystal Chem, Spain) following manufacturer instructions, according to methods of Temple et al (15) and Frieman (16) respectively. Their assay employs sandwich ELISA method. Lipase activity was determined using kinetic colorimetric method following manufacturer instructions (kit, Ref: 1001275, supplied from SPINREACT, Spain). Serum apo-A1and apo-B were determined turbedometricaly (kits of SPINREACT, Spain, Ref.: 1003012 and1003013 respectively) using Anti- Apo A-1 and Anti- Apo B antibodies, following manufacturer instructions.

Statistical analysis

Data were processed and analyzed using the Statistical Package of Social Science version 10.0 (SPSS, version 10.0). The data were expressed as mean ± standard error of mean (Mean±SEM). One-way ANOVA analysis of data was done. A minimum level of significance is considered if P is$\leq$0.05.

RESULTS

In MSG fed pregnant rats, daily food intake and body weight decreased significantly relative to HCC fed rats ($p<0.05$) but non significantly relative to control rats ($p>0.05$) whereas body fat and fat to body weight ratio increased significantly in MSG fed relative to high caloric chow fed and control pregnant rats ($p<0.05$).Moreover, daily food intake, body weight, body fat and fat to body weight ratio increased significantly in HCC fed relative to control pregnant rats ($p<0.05$) (table 1).

The administration of MSG or HCC to pregnant rats leads to a significant induction in Ob gene expression, repression of leptin receptor-b gene expression (figure 1), decrease in lipoprotein lipase activity, increase in blood glucose, serum leptin, apo-B, insulin concentrations, a significant increase in all parameters of lipid profile, relative to those of control preg-

nant rats, except HDL which, in MSG, decreased significantly at 10th day of pregnancy (MSG_{10}) then returned as that of control at 20th day of pregnancy (MSG_{20}) while in HCC it decreased non significantly relative to control rats. Apo-A1 non significantly decreased with high caloric chow (HCC_{10}, HCC_{20}) and MSG20 but significantly decreased with MSG_{10} relative to control groups(table 1).

In offspring of MSG treated rats (PN_{10}-MSG), one and ten days body weight, blood glucose, serum insulin, leptin, TAG, total lipids and ob gene expression were significantly increased relative to those in offspring of control rats (PN_{10}-C) while leptin receptor-b gene expression (figure 2) was significantly decreased relative to offspring of control and HCC rats. On the other hand, offspring of HCC fed rats (PN_{10}-HCC) showed no significant increase in one and ten days body weight, blood glucose, TAG and total lipids whereas ob and leptin receptor-b gene expression (figure 2), serum insulin, and serum leptin were significantly increased relative to those in offspring of control rats (PN_{10}-C) (table 2).

Table (1): Biochemical parameters, ob and leptin receptor-b gene expressions in pregnant rats.

	C_{10} rats	C_{20} rats	HCC_{10} rats	HCC_{20} rats	MSG_{10} rats	MSG_{20} rats
Daily food intake (grams)	4.08±0.48	4.12±0.42	5.8±0.28[a]	6.04±0.61[a]	3.9±0.22	3.92±0.25
Body weight (grams)	236.4±6.5	255.6±7.4	301.6±5.9[a]	309.6±8[a]	237.4±6	245.4±5.3
Body fat (g)	0.36±0.05	0.36±0.04	4.46±0.49[a]	4.5±0.56[a]	5.76±0.39[ab]	5.8±0.5[ab]
Fat to body weight ratio	0.15±0.007	0.14±0.005	1.48±0.08[a]	1.45±0.07[a]	2.42±0.06[ab]	2.37±0.09[ab]
Blood glucose (mg/dl)	84.2±1.2	79.2±1	118.2± 3.2[a]	122.6±2.9[a]	120 ± 2.2[a]	125 ± 2.5[a]
Serum insulin (ng/ml)	13.1±1.2	14.3±1.3	19.4±1.2[a]	20.3±1.4[a]	19.9±1.4[a]	20.2±1.3[a]
Serum leptin (ng/ml)	3.1±0.2	2.8± 0.3	6.5± 0.5[a]	6.2 ±0.6[a]	6.4± 0.6[a]	6.3 ±0.5[a]
LPL (unit/L)	8.6 ± 0.8	8.9 ± 0.9	3.9±0.2[a]	3.7±0.3[a]	3.8±0.1[a]	3.6±0.2[a]
Apo-A1 (mg/dl)	26.2±2.4	28.6±2.6	24.7±3.5	25.6±3.8	19.6±1[a]	22.4±3
Apo-B (mg/dl)	45.1±3.2	42.6±4	62.4±4.3[a]	66.1±4.2[a]	63±4.2[a]	65.5±4.1[a]
TAG (mg/dl)	147.4±13	163.7±15	210.6±4[a]	221.4±5[a]	200.3±5[a]	223.3±3[a]
TCh (mg/dl)	85.2±3	86.5±2	102.7±2[a]	107.5±3[a]	103.2±1[a]	108.6±2[a]
HDLc (mg/dl)	24.5±1.2	26.5±2	23.94±1	24.2±2	21.25±1[a]	24.12±1
LDLc (mg/dl)	31.4±2	27.3±5	42.2±3[a]	38.1±2[a]	41.9±2[a]	39.8±1[a]
VLDLc (mg/dl)	29.4±2.8	32.6±3	41.1 ±2[a]	43.4±1[a]	40.2 ±1[a]	44.6±1[a]
TL (mg/dl)	314.4±26	348.8±15	420.8±5[a]	441.7±6[a]	414.9±4[a]	450.7±3[a]
Ob gene expression (nmol/ml)	3.4±0.45	3.5±0.55	7.7±0.42[a]	7.6±0.82[a]	7.9±0.36[a]	7.8±0.75[a]
Leptin receptor-b gene expression (nmol/ml)	12±1	11.6±1.5	8.12±0.31[a]	8.0±0.65[a]	8.26±0.25[a]	8.1±0.75[a]

a: significant relative to control groups (C_{10} and C_{20}), ($P<0.05$) **b:** significant relative to HCC groups (HCC_{10} and HCC_{20})($P<0.05$), **C_{10}:** control rats at 10th day, **C_{20}:** control rats at 20th day, **HCC_{10}:** high caloric chow rats at 10th day, **HCC_{20}:** high caloric chow rats at 20th day, **MSG_{10}:** monosodium glutamate rats at 10th day, **MSG_{20}:** monosodium glutamate rats at 20th day.

Table(2): Biochemical parameters, ob and leptin receptor-b gene expression in offspring of pregnant rats.

	Offspring of control rats (PN_{10}-C)	Offspring of high caloric chow fed rats (PN_{10}-HCC)	Offspring of MSG treated rats (PN_{10}-MSG)
One day BW (grams)	4.6±0.232	4.7±0.24	7.9±0.17^{ab}
Ten day BW (grams)	15±1	16±2	24.6±1.4^{ab}
Blood glucose (mg/dl)	85±2.5	87±3.5	144±3^{ab}
Serum insulin (ng/ml)	3.7±0.38	5.2±0.45^{a}	5.3±0.2^{a}
Serum leptin (ng/ml)	4.9±0.57	7.1±0.61^{a}	7±0.58^{a}
TAG (mg/dl)	133±8	136±8	182±9$^{a\,b}$
Total lipids (mg/dl)	302.1±11	306±5	378.6±6$^{a\,b}$
Ob gene expression (nmol/ml)	1.8±0.56	3.1±0.34^{a}	3±0.3^{a}
Leptin receptor-b gene expression (nmol/ml)	0.85±0.07	1.15±0.05^{a}	0.55±0.02^{ab}

a: significant relative to PN_{10}-C ($P<0.05$).

b: significant relative to PN_{10}-HCC ($P<0.05$).

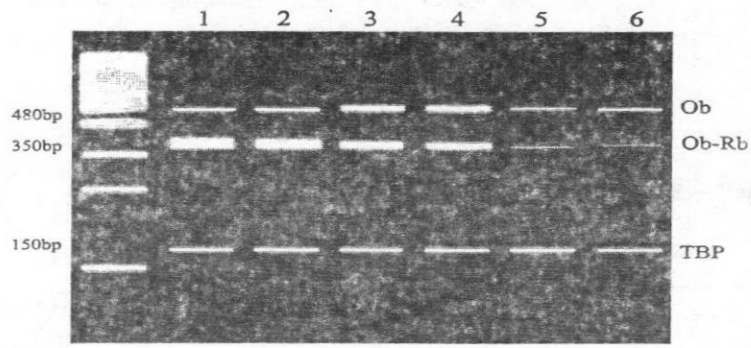

Figure (1): Expression of adipose tissue ob and hypothalamic ob-Rb (leptin receptor-b) mRNA in control and MSG (monosodium glutamate) treated pregnant rats and their offspring.

1 = control pregnant rats sacrificed by decapitation at 10th day of pregnancy.
2 = control pregnant rats sacrificed by decapitation at 20th day of pregnancy.
3 = pregnant rats treated with MSG and sacrificed by decapitation at 10th day of pregnancy.
4 = pregnant rats treated with MSG and sacrificed by decapitation at 20th day of pregnancy.
5 = offspring of control rats (sacrificed by decapitation at 10th post natal day).
6 = offspring of rats treated with MSG (sacrificed by decapitation at 10th post natal day).
TBP = TATA-box binding protein.

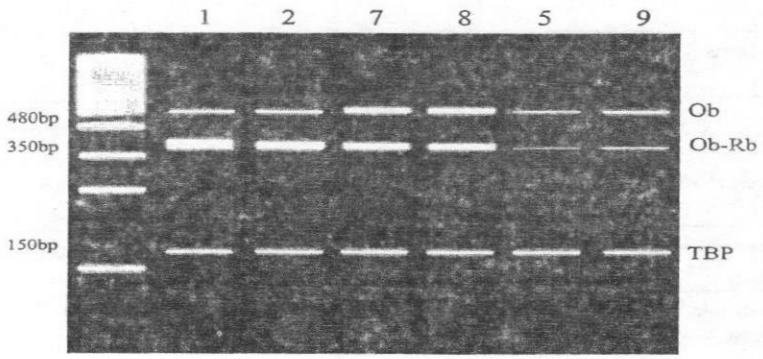

Figure (2): Expression of adipose tissue ob and hypothalamic ob-Rb (leptin receptor-b) mRNA in control and high caloric chow fed pregnant rats and their offspring.

1 = control pregnant rats sacrificed by decapitation at 10th day of pregnancy.
2 = control pregnant rats sacrificed by decapitation at 20th day of pregnancy.
7 = pregnant rats fed high caloric chow and sacrificed by decapitation at 10th day of pregnancy.
8 = pregnant rats fed high caloric chow and sacrificed by decapitation at 20th day of pregnancy.
5 = offspring of control rats (sacrificed by decapitation at 10th post natal day).
9 = offspring of high caloric chow fed rats (sacrificed by decapitation at 10th post natal day).
TBP = TATA-box binding protein.

DISCUSSION

Monosodium glutamate (MSG) is one of the main flavor enhancers used as an ingredient in various food products especially meat. Although MSG was reported to have broad toxic effects on different body organs of human and experimental animals [17], yet it is still widely used in eastern and western diets. Since it is difficult to determine obesity-induced complications in pregnant women, experimental studies are carried out to induce obesity [18]. The model MSG-treated rats is of special interest in the development of obesity which is not a result of overeating [7].

The results of the present study showed that, the body weight and daily food intake in MSG fed pregnant rats were decreased non significantly relative to control pregnant rats (insignificant hypophagia) but significantly relative to high caloric chow pregnant rats, although they had excessively accumulation of subcutaneous, abdominal and gonadal fat and significantly increased fat to body weight ratio comparing with their control and the high caloric chow pregnant rats (table 1).

The relationship between MSG-induced damage of arcuate nucleus and the metabolic changes that produce obesity at mild hypophagia is poorly understood, one of the basic factors is probably the lack of leptin receptors in arcuate nucleus (table1 and figure1). In our study, MSG treatment resulted in massive enhancement of adipose tissue despite an insignificantly lowered food intake and no significant difference in body weight compared with their respective controls. Fat to body weight ratio of MSG10 pregnant rats was about 16.3 times higher than their respective control and 1.63 times higher than high caloric chow pregnant rats. Also it was 16.9 times higher in MSG20 pregnant rats as compared with their controls and 1.63 times as compared with high caloric chow pregnant rats. Our findings are in agreement with previous data on C57BL/6 mice with MSG obesity [8]

The results of the present study showed that the administration of MSG and high caloric chow to pregnant rats leads to significant increase in ob gene expression with decreased leptin receptor-b gene expression (ta-

ble 1 and figure 1) which indicates leptin resistance. Frederich et al [19] observed increased circulating ob protein in MSG mice compared to controls. Defective leptin signaling due to mutations in the leptin gene, obese (ob/ob), of mutations in the receptor (leptin receptor-b) in mice decreased energy expenditure causing a disruption in energy balance which result in obesity [20]. However, in most obese conditions, plasma leptin is elevated well above levels that should suppress food intake and reduce body weight, yet fails to do so [21]. This suggests that a state of leptin resistance is linked to the development of obesity. MSG causes leptin resistance by damaging of arcuate nucleus [22] whereas in models of diet-induced obesity, leptin resistance results from defects influencing leptin transport across the blood brain barrier (BBB) and signaling downstream of the leptin receptor [23]. Current theories further speculate that diet induced leptin resistance may be mediated through blood borne circulating factors associated with high-fat feeding [24].

MSG and HCC fed obese rats develop glucose intolerance as well as insulin resistance to peripheral glucose uptake [25, 26]. This is revealed in table (1) where the increase of serum insulin in pregnant rats administered MSG or HCC is not able to decrease the blood glucose in these rats which indicates the development of insulin resistance. Zorad et al [27] reported that insulin resistance could be due to changes in insulin binding or post receptor insulin effects in target tissues. The stimulatory effect of insulin on glucose transport in adipocytes, cardiac and skeletal muscles decreased due to decreased content of glucose transport protein (GLUT4) and decreased number of insulin receptors [27].

Furthermore, our results demonstrated that administration of MSG or HCC to pregnant rats resulted in an increase in the lipid profile (triglycerides, cholesterol, LDL,VLDL and total lipids) and decrease HDL and LPL(table 1), which suggest increased lipogenesis and decreased lipolysis. Also, apo B increase and apo A1 decrease (table 1) which is an indicator of a cardiac diseases (atherosclerosis). The increase in serum lep-

tin, TAG, cholesterol, total lipids, (table 1), ob gene expression and the decrease in leptin receptor-b gene expression (table 1;figure 1) indicates the development of leptin resistance. All these lead to the development of obesity. These results were in agreement with previous reports [8, 25].

Moreover, the results of the present study showed increased body weight of offspring of MSG treated rats relative to those of the controls or HCC fed pregnant rats (table 2). Campos et al [18] observed an increased percentage of obese rats in the offspring of MSG treated rats compared to the offspring of control rats. This result confirms that maternal obesity, due to MSG, caused obesity in the subsequent generations. There is considerable experimental and clinical evidence that an altered body composition before and during pregnancy, by MSG, produces altered metabolism in the offspring which then have a greater propensity to develop diabetes and/or obesity [28]. However, there is little information about the pathological mechanisms of transgenerational obesity [18]. Also, our results showed that blood glucose was significantly increased in offspring of MSG treated rats relative to those of high caloric chow fed pregnant rats whilst insulin goes up in both which indicates the development of insulin resistance in offspring of MSG treated pregnant rats. Furthermore, our results demonstrated increased serum triacylglycerides (TAG), total lipids (TL), in offspring of MSG treated pregnant rats relative to offspring of control or high caloric chow fed pregnant rats (table 2). In addition, leptin receptor b gene expression was significantly decreased in offspring of MSG treated rats as compared with those of HCC fed pregnant rats whilst serum leptin and ob gene expression go up in both (table 2 and figure 2) which indicates the development of leptin resistance in offspring of pregnant rats treated with MSG. These results were in accord with previous data on high fat fed rats [26].

CONCLUSION

Both MSG and HCC produce obesity in pregnant rats. The MSG induced obesity is manifested by a severe increase in body fat and fat to body weight ratio in spite of hypophagia but no observed changes in body

weight. This might be caused by the leptin resistance resulting from the distractive effect of MSG on leptin receptors (arcuate nucleus). On the other hand, obesity of high caloric chow was manifested by an increase of body weight, fat and fat to body weight ratio as a result of hyperphagia with consequence leptin resistance in pregnant rats but not in their offspring. Both models are accompanied with a worse metabolic changes in insulin, glucose, lipid profile but MSG model was the worst. Moreover, maternal obesity in pregnancy, caused by MSG, has a greater impact on offspring metabolism and body weight than that induced by high calorie chow feeds alone.

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الملخص العربى

دور أحادى صوديوم الجلوتامات والطعام الغنى بالسعرات الحرارية فى السمنة فى الفئران الحوامل و أبنائهم

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أجرى هذا البحث لدراسة دور أحادى صوديوم الجلوتامات والطعام الغنى بالسعرات الحرارية فى السمنة فى الفئران الحوامل وأبنائهم وتأثير أحادى صوديوم الجلوتامات والطعام الغنى بالسعرات الحرارية على بعض مؤشرات أمراض القلب وقد تم إجراء هذا البحث على ٩٠ من إناث فئران الألبينوتزن ١٥٠ جرام تقريباً وتم تقسيم هذه الفئران إلى ثلاث مجموعات:

١- **المجموعة الأولى (الضابطة)** : وتشمل ٣٠فأر وتم تغذيتها بطعام قياسى لمدة ٣ شهور وتم إعتبارهم كمجموعة ضابطة .

٢- **المجموعة الثانية (مجموعة أحادى صوديوم الجلوتامات)** : و تشمل ٣٠فأر وتم تغذيتها بطعام قياسى مضافاً إليه أحادى صوديوم الجلوتامات بجرعة ١٠٠ ملليجرام / كيلو جرام من وزن الفأر لمدة ٣ شهور .

٣- **المجموعة الثالثة (مجموعة الطعام الغنى بالسعرات الحرارية)** : وتشمل ٣٠فأر وتم تغذيتها بطعام غنى بالسعرات الحرارية لمدة ٣ شهور .

وقد تزاوجت هذه الفئران و أصبحت حوامل ثم تم تقسيمها إلى :

١- **المجموعة الأولى أ** : وتشمل ١٠ من إناث فئران الألبينو الحوامل التى لم يضاف لها فى الغذاء أحادى صوديوم الجلوتامات أو اى طعام غنى بالسعرات الحرارية (المجموعة الضابطة) و قد تم ذبح هذه الفئران بعد ١٠ أيام من الحمل.

٢- **المجموعة الأولى ب** : وتشمل ١٠ من إناث فئران الألبينو الحوامل التى لم يضاف لها فى الغذاء أحادى صوديوم الجلوتامات أو اى طعام غنى بالسعرات الحرارية (المجموعة الضابطة) وقد تم ذبح هذه الفئران بعد ٢٠ يوماً من الحمل.

٣- **المجموعة الأولى ج** : وتشمل ١٠ من إناث فئران الألبينو الحوامل التى لم يضاف لها فى الغذاء أحادى صوديوم الجلوتامات أو اى طعام غنى بالسعرات الحرارية وقد تم ترك هذه الفئران حتى نهاية الحمل و بعد الولادة تم ذبح الفئران الحديثة الولادة عند اليوم العاشر من عمرها.

٤- **المجموعة الثانية أ** : وتشمل ١٠ من إناث فئران الألبينو الحوامل التى أضيف لها فى الغذاء أحادى صوديوم الجلوتامات وقد تم ذبح هذه الفئران بعد ١٠ أيام من الحمل.

٥- **المجموعة الثانية ب** : وتشمل ١٠ من إناث فئران الألبينو الحوامل التى أضيف لها فى الغذاء أحادى صوديوم الجلوتامات وقد تم ذبح هذه الفئران بعد ٢٠ يوما من الحمل.

٦- **المجموعة الثانية ج** : وتشمل ١٠ من إناث فئران الألبينو الحوامل التى أضيف لها فى الغذاء أحادى صوديوم الجلوتامات وقد تم ترك هذه حتى نهاية الحمل وبعد الولادة تم ذبح الفئران الحديثة الولادة عند اليوم العاشر من عمرها.

٧- **المجموعة الثالثة أ** : و تشمل ١٠ من إناث فئران الألبينو الحوامل التى تم تغذيتها بالطعام الغنى بالسعرات الحرارية وقد تم ذبح هذه الفئران بعد ١٠ أيام من الحمل.

٨- **المجموعة الثالثة ب** : وتشمل ١٠ من إناث فئران الألبينو الحوامل التى تم تغذيتها بالطعام الغنى بالسعرات الحرارية وقد تم ذبح هذه الفئران بعد ٢٠ يوماً من الحمل.

٩- **المجموعة الثالثة ج** : وتشمل ١٠ من إناث فئران الألبينو الحوامل التى تم تغذيتها بالطعام الغنى بالسعرات الحرارية وقد تم ترك هذه الفئران حتى نهاية الحمل وبعد الولادة تم ذبح الفئران الحديثة الولادة عند اليوم العاشر من عمرها.

و قد تم أخذ عينات الدم من كل الفئران عند ذبحها و ذلك لقياس مستوى الجلوكوز والأنسولين

و الدهون والكوليستيرول فى الدم وأيضاً تم أخذ عينات من المخ والدهون وتم حفظها فى النيتروجين السائل لتحديد ودراسة چينات السمنة وچينات مستقبلات هرمون اللبتين.

ولقد أظهر البحث أن تناول أحادى صوديوم الجلوتامات أو الطعام الغنى بالسعرات الحرارية بكثرة يعتبر من الأسباب الرئيسية للسمنة و يزيد من خطورة حدوث تصلب الشرايين. كما أظهر هذا البحث أيضاً أن السمنة أثناء الحمل لها خطورة كبيرة على صحة الأم و الطفل .

ونستخلص من هذا البحث خطورة الإفراط فى تناول الإضافات الغذائية مثل أحادى صوديوم الجلوتامات أو الطعام الغنى بالسعرات الحرارية على الصحة وبخاصة أنه يسبب حدوث الأمراض مثل السمنة و تصلب الشرايين. ونستخلص أيضا أن الوقاية من السمنة أثناء الحمل مهم للحفاظ على صحة الأم والطفل .